

What Is Claimed Is:

1. A composition, comprising a crystal of isolated *Streptococcus pneumoniae* acyl carrier protein synthase.

2. The composition of claim 1, wherein said crystal effectively diffracts X-rays, and permits the determination of the atomic coordinates of said acyl carrier protein synthase to a resolution of about 2.0 Å.

3. The composition of claim 1, wherein said *Streptococcus pneumoniae* acyl carrier protein synthase has an active site cavity comprising the 3',5'-adenosine diphosphate binding site shown in Figure 9.

4. The composition of claim 1, wherein said *Streptococcus pneumoniae* acyl carrier protein synthase is a homotrimer, wherein each protomer comprises the following structural motifs:

- (a) a three-stranded anti-parallel β -sheet formed by strands $\beta 1$, $\beta 5$, and $\beta 4$;
- (b) a long α -helix that packs diagonally against said β -sheet, together with α -helices $\alpha 1$, $\alpha 2$, $\alpha 3$, and $\alpha 4$ of an anti-parallel four helical bundle; and
- (c) a long, extended loop with a two-strand anti-parallel β -sheet comprising strands $\beta 2$ and $\beta 3$,

wherein said structural motifs (a), (b), and (c) are organized such that said long helix $\alpha 4$ runs through said homotrimer, and is surrounded by the remainder of said structural motifs, as shown in Figures 8(B) and 8(C) .

5. The composition of claim 1, wherein said crystal belongs to orthorhombic space group $P2_12_12_1$, with unit cell dimensions of $a = 49.8 \text{ \AA}$, $b = 59.6 \text{ \AA}$, and $c = 114.7 \text{ \AA}$, or monoclinic space group $C2$, with unit cell dimensions of $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, and $\beta = 98.7^\circ$.

6. The composition of claim 1, wherein said acyl carrier protein synthase comprises selenocysteine or selenomethionine.

7. The composition of claim 1, wherein said acyl carrier protein synthase comprises a heavy metal atom.

8. The composition of claim 1, further comprising a chemical compound complexed covalently or non-covalently with said crystal.

9. The composition of claim 8, wherein said chemical compound is 3',5'-adenosine diphosphate.

10. The composition claim 9, wherein said crystal belongs to monoclinic space group $C2$, with unit cell dimensions of $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, and $\beta = 98.7^\circ$.

11. The composition of claim 1, wherein said *Streptococcus pneumoniae* acyl carrier protein synthase has the amino acid sequence shown in SEQ ID NO:1.

12. The composition of claim 1, wherein said crystal has the atomic coordinates shown in Table 3 or Table 4.

13. The composition of claim 9, wherein said crystal has the atomic coordinates shown in Table 5.

14. A composition, comprising a crystal of *Streptococcus pneumoniae* acyl carrier protein synthase having the amino acid sequence shown in SEQ ID NO:1 wherein methionine is substituted with selenomethionine,

wherein said acyl carrier protein synthase is a homotrimer, wherein each protomer comprises the following structural motifs:

- (d) a three-stranded anti-parallel β -sheet formed by strands $\beta 1$, $\beta 5$, and $\beta 4$;
- (e) a long α -helix that packs diagonally against said β -sheet, together with α -helices $\alpha 1$, $\alpha 2$, $\alpha 3$, and $\alpha 4$ of an anti-parallel four helical bundle; and
- (f) a long, extended loop with a two-strand anti-parallel β -sheet comprising strands $\beta 2$ and $\beta 3$,

wherein said structural motifs (a), (b), and (c) are organized such that said long helix $\alpha 4$ runs through said homotrimer, and is surrounded by the remainder of said structural motifs, as shown in Figures 8(B) and 8(C);

wherein when said acyl carrier protein synthase is in native form, said crystal belongs to orthorhombic space group $P2_12_12_1$, with unit cell dimensions of $a = 49.8 \text{ \AA}$, $b = 59.6 \text{ \AA}$, and $c = 114.7 \text{ \AA}$, or monoclinic space group $C2$, with unit cell dimensions of $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, and $\beta = 98.7^\circ$, and wherein said crystal has the atomic coordinates shown in Table 3 or Table 4, respectively; and

wherein when said acyl carrier protein synthase is complexed with 3',5'-adenosine diphosphate, said crystal belongs to monoclinic space group $C2$, with unit cell dimensions of $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, and $\beta = 98.7^\circ$, and wherein said crystal has the atomic coordinates shown in Table 5.

15. An enzyme active site crystal structure comprising the 3',5'-adenosine diphosphate binding site shown in Figure 9.

16. The enzyme active site crystal structure of claim 15, wherein said enzyme comprises isolated, properly folded *Streptococcus pneumoniae* acyl carrier protein synthase, or a fragment thereof comprising said active site.

17. A method of isolating *Streptococcus pneumoniae* acyl carrier protein synthase, comprising:

- (a) growing said *Streptococcus pneumoniae* in a medium lacking methionine but containing L-selenomethionine;
- (b) preparing a cell extract of said *Streptococcus pneumoniae*;
- (c) centrifuging said cell extract to produce a supernatant fraction, and collecting said supernatant fraction;
- (d) chromatographing said supernatant fraction on a cation exchange column in buffer containing dithiothreitol or β -mercaptoethanol, and collecting fractions containing said *Streptococcus pneumoniae* acyl carrier protein synthase;
- (e) chromatographing said fractions of step (d) on a gel filtration column in buffer containing dithiothreitol or β -mercaptoethanol, and collecting fractions containing said *Streptococcus pneumoniae* acyl carrier protein synthase comprising L-selenomethionine.

18. The method of claim 17, further comprising chromatographing said fractions of step (e) on an anion exchange column in buffer containing dithiothreitol or β -mercaptoethanol, and collecting fractions containing said *Streptococcus pneumoniae* acyl carrier protein synthase.

19. The method of claim 18, wherein said *Streptococcus pneumoniae* acyl carrier protein synthase has the amino acid sequence shown in SEQ ID NO:1, wherein methionine is replaced with L-selenomethionine.

20. Isolated *Streptococcus pneumoniae* acyl carrier protein synthase produced by the method of claim 19.

21. A method of producing a crystal of *Streptococcus pneumoniae* acyl carrier protein synthase that diffracts X-rays, comprising:

- (a) providing *Streptococcus pneumoniae* acyl carrier protein synthase isolated according to claim 19;
- (b) concentrating said acyl carrier protein synthase to 8 mg/ml in a solution containing 10 mM MgCl₂, 14 mM KCl, and 20 mM Tris-HCl at pH 7.1 to produce a concentrated protein solution;
- (c) equilibrating a 4 µl drop of said acyl carrier protein synthase in a solution comprising a mixture of 1:1, v/v, concentrated protein solution as in step (b)/reservoir solution over a 500 µl reservoir solution comprising 8-15% polyethyleneglycol 4000, 200 mM ammonium sulfate, and 100 mM citrate buffer at pH 4.5; and
- (d) growing a crystal of said acyl carrier protein synthase by vapor diffusion at 294K for at least 4 to 5 days.

22. The method of claim 21, further comprising determining a three-dimensional structure of said crystal.

23. The method of claim 21, wherein said crystal belongs to orthorhombic space group $P2_12_12_1$, having unit cell parameters $a = 49.8 \text{ \AA}$, $b = 59.6 \text{ \AA}$, $c = 114.7 \text{ \AA}$, or monoclinic space group $C2$, having unit cell parameters $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, $\beta = 98.7^\circ$), comprises one homotrimeric molecule per asymmetric unit, and has the atomic coordinates shown in Table 3 or Table 4.

24. The method of claim 21, further comprising testing the ability of a compound to form a complex with an active site of said acyl carrier protein synthase by including said compound in said concentrated protein solution of step (b).

5 25. The method of claim 21, further comprising contacting said crystal of acyl carrier protein synthase and a solution comprising a compound of interest to form a mixture, incubating said mixture to permit said compound to diffuse into said crystal, and determining whether said compound forms a complex with said acyl carrier protein synthase.

10 26. The method of claim 25, wherein said compound is 3',5'-adenosine diphosphate.

15 27. The method of claim 25, wherein when said compound forms a complex with said acyl carrier protein synthase, determining a three-dimensional structure of said acyl carrier protein synthase/compound crystal complex.

20 28. The method of claim 26, wherein said crystal belongs to monoclinic space group *C*2, having unit cell parameters $a = 120.2 \text{ \AA}$, $b = 62.3 \text{ \AA}$, $c = 51.7 \text{ \AA}$, $\beta = 98.7^\circ$, comprises one homotrimeric molecule per asymmetric unit, and has the atomic coordinates shown in Table 5.

25 29. A crystal of *Streptococcus pneumoniae* acyl carrier protein synthase produced by the method of claim 21.

30 30. A crystal of *Streptococcus pneumoniae* acyl carrier protein synthase produced by the method of claim 24.

31. A crystal of *Streptococcus pneumoniae* acyl carrier protein synthase produced by the method of claim 25.

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